

WE CLAIM:

1. A method of inducing at least one site-directed double-strand break in DNA of a cell, said method comprising
 - (a) providing cells containing double-stranded DNA, wherein said DNA comprises at least one I-Sce I restriction site;
 - (b) transfecting said cells with at least a plasmid comprising DNA encoding the I-Sce I meganuclease; and
 - (c) selecting cells in which at least one double-strand break has been induced.
2. The method of claim 1, wherein said cell is selected from the group consisting of a mammalian cell, a yeast cell, and a plant cell.
3. The method of claim 2, wherein said cell is an NIH3T3 cell containing the G-MtkPL virus.
4. The method of claim 1, wherein said plasmid is pCMV(I-Sce I+).
5. A method of inducing homologous recombination between chromosomal DNA of a cell and exogenous DNA added to said cell, said method comprising
 - (a) providing cells containing chromosomal DNA, wherein said DNA comprises at least one I-Sce I restriction site;
 - (b) transfecting said cells with a plasmid comprising exogenous DNA, and with a plasmid comprising DNA encoding the I-Sce I meganuclease; and
 - (c) selecting cells in which said exogenous DNA is inserted into said chromosomal DNA.

6. The method of claim 5, wherein said cell is selected from the group consisting of a mammalian cell, a yeast cell, and a plant cell.

7. The method of claim 6, said cell is an NIH3T3 cell containing the G-MtkPL virus.

8. The method of claim 5, wherein said plasmid is pCMV(I-Sce I+).

9. A method of inducing homologous recombination between chromosomal DNA of a cell and exogenous DNA added to said cell, said method comprising

- (a) providing cells comprising chromosomal DNA;
- (b) inserting at least one I-Sce I restriction site in said chromosomal DNA;
- (c) transfecting said cells with a first plasmid comprising exogenous DNA, and with a second plasmid comprising DNA encoding the I-Sce I meganuclease; and
- (d) selecting cells in which said exogenous DNA is inserted into said chromosomal DNA.

10. The method of claim 9, wherein said cell is selected from the group consisting of a mammalian cell, a yeast cell, and a plant cell.

11. The method of claim 9, wherein said first plasmid is pCMV(I-Sce I+).

12. The method of claim 9, wherein said second plasmid is pVRneo.

13. A method of inducing at least one site-directed break in DNA of a cell and inserting DNA encoding a polypeptide, said method comprising,

- (a) providing cells containing double-stranded DNA, wherein said cells are capable of being transformed by a DNA comprising a I-Sce I restriction site and DNA encoding said polypeptide;
- (b) adding *Sce I* enzyme or transforming said cell with DNA encoding *Sce I* enzyme;
- (c) transfecting said cells with said DNA encoding said polypeptide or with a vector containing said DNA; and
- (d) selecting cells transfected with said DNA or said vector, wherein said cells express said polypeptide.

14. A recombinant eukaryotic cell transformed by the method of any one of claims 1 and 13.

15. A transgenic animal comprising a cell transformed by the method of any one of claims 1 and 13.

16. A method of expressing a polypeptide in a transgenic animal, said method comprising transforming embryonic stem cells with a DNA comprising a I-Sce I restriction site and DNA encoding said polypeptide, and detecting expression of said polypeptide in a transgenic animal resulting from said transformed embryonic stem cells.

17. A recombinant stem cell expressing a polypeptide, wherein said stem cell is transformed by a DNA comprising a I-Sce I restriction site and DNA encoding said polypeptide by

- (a) adding *Sce I* enzyme to said cell or transforming said cell with a vector containing the gene coding for *Sce I* enzyme;
- (b) transfecting said cells with said DNA encoding said polypeptide; and
- (c) selecting cells transfected with said DNA, wherein said cells express said polypeptide.

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18. A recombinant eukaryotic cell as claimed in any one of claims 4 and 7 wherein said polypeptide is a foreign antigen to the cell.

19. The recombinant eukaryotic cell as claimed in claim 14 wherein cell is a mammalian cell line.

20. The recombinant eukaryotic cell as claimed in claim 14 wherein cell is a yeast.

21. A method of inducing at least one site-directed break in DNA of cells and inserting DNA encoding a polypeptide, wherein said cells express at least one protein product, said method comprising,

(a) providing cells containing double-stranded DNA, wherein said cells are capable of being transformed by a DNA comprising a *I-Sce I* restriction site and DNA encoding said polypeptide;

(b) adding *Sce I* enzyme to said cells or transforming said cells with DNA encoding *Sce I* enzyme;

(c) transfecting said cells with said DNA encoding said polypeptide or with a vector containing said DNA; and

(d) selecting cells transfected with said DNA or said vector, wherein said cells express said polypeptide and do not express said protein product.

22. A recombinant cell transformed by the method of claim 21.

ADD
A3 add E1